B.7.6 Residues Resulting from Supervised Trials (Annex IIA 6.3; Annex IIIA 8.3)

B.7.6.1 Residues in Target Crops

B.7.6.1.1 Globe artichoke

Document ID:	MRID No. 50489801 PMRA No. 2876292
Report:	Dorschner, K. (2017) "Sulfoxaflor: Magnitude of the Residue on Artichoke (Globe)". IR-4 PR No. 10858. Laboratory Identification Number 10858.14-FLR02. Unpublished study prepared by IR-4 Project. Rutgers, The State University of New Jersey. North Princeton, NJ, USA. 162 pages.
Guidelines:	EPA OCSPP Harmonized Test Guideline 860.1500 Crop Field Trials (August 1996) PMRA Regulatory Directive DIR98-02 – Residue Chemistry Guidelines, Section 9 – Crop Field Trials PMRA Regulatory Directive DIR2010-05 – Revisions to the Residue Chemistry Crop Field Trial Requirements OECD Guideline 509 Crop Field Trial (September 2009)
GLP Compliance:	No deviations from regulatory requirements were reported which would have an impact on the validity of the study.
Acceptability:	The study is considered scientifically acceptable.
Evaluator:	Jack Giordano, Chemist, RAB2/HED (7509P)

EXECUTIVE SUMMARY

Four field trials for sulfoxaflor on globe artichokes were conducted in Canada and the United States encompassing North American Free Trade Agreement (NAFTA) Growing Regions 5B (1 trial in Quebec) and 10 (3 trials in California) during the 2014 growing season. Two field trials were attempted in British Columbia (Zone 12) but both trials failed because of poor flowering. All trials except sites CA64 and CA66 meet the "Criteria for Determining Independence of Crop Field Trials" (November 2014) and are considered independent. As such, there are a total of 3 independent field trials for sulfoxaflor on globe artichokes.

At each trial location, sulfoxaflor, formulated as a suspension concentrate (Closer SC, EPA Reg. 62719-623 and PCP # 30826), was applied to globe artichokes as three foliar directed or broadcast applications at rates of 0.088-0.092 lbs a.i./A (98.2-103.7 g a.i./ha). The re-treatment intervals were 6-9 days, and the total application rates were 0.269-0.272 lbs a.i./A (301.1-306.4 g a.i./ha). An adjuvant was added to the spray mixture for all applications. Globe artichokes were harvested at a preharvest interval (PHI) of 3 days. In one trial (CA64), samples were collected at different time intervals (PHIs of 1, 7, 14, and 20 days) to monitor residue decline.

All samples were maintained frozen at the testing facility, during shipping to the laboratory, and were stored frozen until analysis. The maximum storage interval for samples between harvest and extraction was 778 days (~26 months). Residues of sulfoxaflor have been shown to be stable in globe artichokes for up to 735 days (24.5 months) under frozen conditions. Adequate

storage stability data are therefore available to support the storage conditions and intervals for samples in the current trials.

Samples in the current study were analyzed using Method 091116, an LC/MS/MS method to determine residues of sulfoxaflor (XDE-208) and metabolites X11719474 and X11721061. Acceptable method validation and concurrent recoveries were reported for globe artichoke samples at fortification levels of 0.01-2.0 mg/kg (ppm), thus validating the method. The limit of quantitation (LOQ), based on the lowest level of method validation, was 0.01 ppm per analyte for globe artichokes.

Individual sample (and per-trial average) combined sulfoxaflor, X11719474, and X11721061 residues in globe artichoke ranged from <0.177 ppm to <0.337 ppm (<0.248 ppm to <0.330 ppm). Residue decline data show that combined residues of sulfoxaflor, X11719474, and X11921061 decrease in globe artichokes with increasing PHIs.

I. MATERIALS AND METHODS

A. MATERIALS

Table B.7.6.1.1-1. Nomenclature for Sulfoxaflor and Metabolites of Interest.	
Common name	Sulfoxaflor
Identity	*N*-[methyloxido[1-[6-(trifluoromethyl)-3-pyridinyl]ethyl]-λ^4-sulfanylidene]cyanamide
CAS no.	946578-00-3
Company experimental name	XDE-208 (Dow Agro) ASF 1069 (Syngenta)
Metabolite	X11719474
Identity	*N*-((methyl)oxido)(1-[6-(trifluoromethyl)pyridine-3-yl]ethyl)-λ^4-sulfanylidene)urea
Metabolite	X11721061
Identity	1-[6-(trifluoromethyl)pyridine-3-yl]ethanol

B. Study Design

1. Test Procedure

A total of 4 residue trials in/on globe artichokes were conducted with a suspension concentrate of sulfoxaflor (Closer SC) during the 2014 growing season (Table B.7.6.1.1-2A).

Table B.7.6.1.1-2A. Trial Numbers and Geographical Locations.

Crop	Region														Total
	1	2	3	4	5/5A/5B	6	7	8	9	10	11	12	13	14	
Globe artichokes	--	--	--	--	1	--	--	--	--	2[1]	--	--	--	--	3

[1] Although 3 trials were conducted in Zone 10, trial sites CA64 and CA66 are considered to be dependent (Table B.7.6.1.1-2B).

Table B.7.6.1.1-2B. Independent Trial Determination[1]			
Crop	Trial Nos.	Differences	Decision
Globe artichoke	CA64 and CA66	Independently prepared tank mixes at each site: The tank mix composition used at each site is different. As such independently prepared tank mixes were used at each site. Location: Both trials were conducted in Salinas, CA. Timing: The applications at each trial site were made less than 30 days apart. Variety: Both trials used the $F_1$41 annual variety of globe artichokes.	Dependent due to same location of trials, similar application dates, and same variety.

[1] All assessments are based on the replicate trial guidance presented in draft memo 568_Criteria for Independence of Trials 04/23/2013 (EPA) and final memo Criteria for Independence of Crop Field Trials November 2014 (PMRA).

Locations and detailed use patterns for the trials are provided in Table B.7.6.1.1-3.

Table B.7.6.1.1-3. Study Use Pattern.							
Location: City, State/Province; Year (Trial ID)[1]	End-use Product/ Formulation (% a.i.)	Method of Application/ Timing of Application	Volume (gal/A) [L/ha]	Rate per Application (lbs a.i./A) [g a.i./ha]	Retreatment Interval (days)	Total Rate (lbs a.i./A) [g a.i./ha]	Surfactant/ Adjuvant
Salinas, CA; 2014 (CA64)	Closer SC/ Suspension concentrate (2 lbs a.i./gal) [240 g a.i./L]	1. Foliar directed/ Producing	10 [94]	0.091 [101.7]	--	0.272 [304.4]	Exit NIS
		2. Foliar directed/ Producing	10 [94]	0.091 [101.6]	7		
		3. Foliar directed/ Producing	10 [94]	0.09 [101.2]	6		
Castroville, CA; 2014 (CA65)	Closer SC/ Suspension concentrate (2 lbs a.i./gal) [240 g a.i./L]	1. Foliar directed/ Producing	75 [705]	0.090 [101.4]	--	0.272 [306.4]	Exit NIS
		2. Foliar directed/ Producing	75 [704]	0.090 [101.3]	7		
		3. Foliar directed/ Producing	77 [721]	0.092 [103.7]	9		
Salinas, CA; 2014 (CA66)	Closer SC/ Suspension concentrate (2 lbs a.i./gal) [240 g a.i./L]	1. Foliar directed/ Producing	100 [931]	0.090 [100.5]	--	0.269 [301.1]	Exit NIS
		2. Foliar directed/ Producing	99 [930]	0.090 [100.4]	9		
		3. Foliar directed/ Producing	99 [929]	0.089 [100.3]	8		
L'Acadie, QC; 2014 (QC419)	Closer SC/ Suspension concentrate (2 lbs a.i./gal) [240 g a.i./L]	1. Foliar broadcast/ Buds at 40%	53 [493]	0.091 [101.6]	--	0.271 [302.5]	Agral 90 NIS
		2. Foliar broadcast/ Buds at 60%	51 [477]	0.088 [98.2]	8		
		3. Foliar broadcast/ Buds at 60%	53 [498]	0.092 [102.6]	7		

[1] All Trial ID #s have the prefix 10858.14-

Globe artichokes were grown and maintained according to typical agricultural practices. Irrigation was used. No unusual weather conditions were reported during the study.

Sample Handling and Preparation

Sampling started in the untreated control plot and ended in the treated plot. Globe artichoke flower heads were harvested using artichoke knives and collected from plants located throughout all rows of each plot. One plant at row ends were avoided. At trial site CA65, 80% of the plot was treated at a much higher rate due to an error in the tank mix preparation. The remaining 20% of the field was treated the following day at the appropriate application rate, and samples were collected only from this portion of the plot. All samples were placed into frozen storage within

35 minutes of harvest. The samples were shipped to the analytical laboratory (IR-4 Southern Region Laboratory in Gainesville, Florida) frozen by ACDS freezer truck. All samples arrived frozen and intact at the analytical laboratory. The samples were checked in, ground with dry ice, and then stored frozen (< -20°C) until extraction and analysis.

2. Description of Analytical Procedures

Samples of globe artichokes were analyzed for residues of sulfoxaflor, X11719474, and X11721061 using a working method very similar to the reference Method 091116, entitled: "Enforcement Method for the Determination of Sulfoxaflor (XDE-208) and its Main Metabolites in Agricultural Commodities using Offline Solid-Phase Extraction and Liquid Chromatography with Tandem Mass Spectrometry Detection" (PMRA # 1941241). The reference method has been reviewed by PMRA (PMRA # 2313516) and was deemed acceptable for enforcement. Minor modifications were made to the reference method to improve performance, such as centrifuging samples for 5 minutes instead of 3 minutes, making up the mixed internal standard solution in acetonitrile instead of methanol/glycerin, and adding 2.5 ml of sample to the centrifuge tube instead of 500 µl. The sample evaporator was at 60°C instead of 40°C, and the sample was concentrated to about 0.5 ml instead of to near dryness. A Supelclean ENVI-Carb SPE tube was used instead of an Oasis HLB 96 well-plate. The sample was concentrated to reduce the amount of acetonitrile and brought up to the 5 ml mark with 5/95 acetonitrile/water and 0.1% formic acid. The extract was filtered prior to analysis, and different LC/MS/MS model, HPLC column, mobile phases, gradient, and injection volume were used. Finally, negative ionization was used for sulfoxaflor and its internal standard X11843864 instead of positive ionization, as negative ionization provides increased sensitivity and lower background.

Briefly, samples were extracted by homogenizing and shaking with 80/20 acetonitrile/water. Extracts were centrifuged, and then an aliquot of the extract was combined with internal standard solution and evaporated. The extracts requiring dilution were diluted before the addition of internal standard. Aqueous sodium hydroxide was added and hydrolyzed at 50°C. The extracts were acidified with aqueous formic acid and incubated at 50°C with glucosidase from Aspergillus Niger solution. The solution was purified with a Supelclean ENVI-Carb SPE cartridge. The extracts were filtered before analysis using LC/MS/MS with heated electro-spray ionization. The LOQ was 0.01 ppm for each analyte, based on the lowest level of method validation.

II. RESULTS AND DISCUSSION

Method performance was evaluated during method validation and by use of concurrent recovery samples by fortifying globe artichokes flower heads at 0.01 ppm (n=6), 0.1 ppm (n=3), and 2.0 ppm (n=4) of each analyte. All recoveries were within the acceptable range of 70% to 120% (Table B.7.6.1.1-4); therefore, the method was considered valid for the analysis of sulfoxaflor, X11719474, and X11721061 residues in globe artichoke matrices. The fortification levels did bracket the measured residues.

Table B.7.6.1.1-4. Summary of Method Validation and Concurrent Recoveries of Sulfoxaflor from Asparagus.			
Matrix	Fortification Level (ppm)	Recoveries (%)	Mean ± Std. Dev. (%)
Sulfoxaflor			
Globe artichokes (Method validation)	0.01	106, 107, 106	106 ± 0.58
	0.1	83, 86, 84	84 ± 1.5
	2.0	84, 84, 86	85 ± 1.2
Globe artichokes (Concurrent recovery)	0.01	105, 113, 111	110 ± 4.2
	2.0	89	--
Metabolite X11719474			
Globe artichokes (Method validation)	0.01	98, 96, 96	97 ± 1.2
	0.1	81, 83, 82	82 ± 1.0
	2.0	82, 83, 83	83 ± 0.6
Globe artichokes (Concurrent recovery)	0.01	96, 101, 100	99 ± 2.6
	2.0	88	--
Metabolite X11721061			
Globe artichokes (Method validation)	0.01	93, 91, 92	92 ± 1
	0.1	81, 85, 82	83 ± 2.1
	2.0	84, 84, 86	85 ± 1.2
Globe artichokes (Concurrent recovery)	0.01	87, 94, 93	91 ± 3.8
	2.0	92	--

The detector response was linear (coefficient of determination, r^2 >0.99) within the range of 0.05 ng/ml to 5.0 ng/ml. Representative chromatograms of control samples, fortified samples and treated samples were provided. The control chromatograms generally had no peaks of interest above the chromatographic background. The fortified sample chromatograms contained only the analyte of interest, and peaks were symmetrical and well defined. Metabolites were expressed in parent equivalents.

The field residue samples were stored frozen a maximum of 778 days (~26 months) from harvest to extraction (Table B.7.6.1.1-5A). Residues were determined within 7 days of extraction.

Freezer storage stability data were generated concurrently with the globe artichoke field trials (Table B.7.6.1.1-5B). Globe artichoke samples were fortified with 0.1 ppm of sulfoxaflor, X11719474, and X11721061 and stored frozen for 735 days (24.5 months). Freezer storage stability recoveries (corrected for concurrent recoveries) were 107%, 99%, and 163% for sulfoxaflor, X11719474, and X11721061, respectively. Although samples were stored for 1.5 months longer than the concurrent storage stability study, given that good recoveries were observed after 735 days of frozen storage, it is not anticipated that residues in globe artichoke matrices would have degraded below acceptable levels during the additional storage sample time. Therefore it is expected that sulfoxaflor, X11719474, and X11721061 residues were stable in globe artichokes under frozen storage for the duration of the storage period.

Table B.7.6.1.1-5A. Summary of Storage Conditions.			
Matrix	Storage Temperature (°C)	Actual Storage Duration[1] (days/months)	Interval of Demonstrated Storage Stability (days/months)
Flower heads (Globe artichoke)	<-20	778 days (~26 months)	A concurrent freezer storage stability study was conducted. The data showed that sulfoxaflor, X11719474, and X11721061 residues are stable when stored frozen in globe artichokes for 735 days (24.5 months) (Table B.7.6.1.1-5B). Therefore, there are no concerns regarding residue stability in storage in this study.

[1] From harvest to residue extraction. Residues were determined within 7 days of extraction.

Table B.7.6.1.1-5B. Concurrent Freezer Storage Stability Study.						
Matrix	Analyte	Storage Period (days)	Fortification Level (ppm)	Freezer Storage Recovery (%) [Average Recovery]	Concurrent Recovery (%)	Corrected Freezer Storage Recovery[1] (%)
Flower heads (Globe artichoke)	Sulfoxaflor	735 (24.5 months)	0.1	90, 89, 91 [90]	84	107
	X11719474		0.1	80, 82, 84 [82]	83	99
	X11721061		0.1	134, 134, 138 [135]	83	163

[1] Corrected for recoveries <100% using the following: (Average Freezer Storage Recovery/Concurrent Recovery)*100

The results from these trials showed that when harvested 3 days after the last of 3 applications at a seasonal rate of 0.269-0.272 lbs a.i./A (301.1-304.9 g a.i./ha), average combined residues of sulfoxaflor, X11719474, and X11721061 in globe artichoke flower heads ranged from <0.204 ppm to <0.311 ppm (Tables B.7.6.1.1-6 and B.7.6.1.1-7).

In the residue decline trials, mean residue level decreased from <0.329 ppm to <0.0493 ppm in globe artichoke flower heads between PHIs of 1 and 20 days.

Table B.7.6.1.1-6. Residue Data from Globe Artichoke Field Trials with Sulfoxaflor.										
Location: City, State/Province; Year (Trial ID)	Region	Crop/ Variety	Matrix	End-Use Product	Rate (lbs a.i./A) [g a.i./ha]	PHI (days)	Residues[1,2] (ppm)			
							Sulfoxaflor	X11719474	X11721061	Total[3] (per-trial average)
Salinas, CA; 2014 (CA64 and CA66)	10	Artichoke/ F1 41 Annual	Flower head	Closer SC	0.269-0.272 [301.1-304.4]	1	0.291	<0.01	0.0538	<0.355, <0.304 (<0.329)
							0.251	<0.01	0.0429	
						3	0.254	<0.01	0.0630	<0.327, <0.256, <0.232, <0.177 (<0.248)
							0.197	<0.01	0.0490	
							0.149	<0.01	0.0727	
							0.114	<0.01	0.0532	
						7	0.127	<0.01	0.0667	<0.204, <0.349 (<0.276)
							0.258	<0.01	0.0809	
						14	0.0421	<0.01	0.0402	<0.0923, <0.126 (<0.109)
							0.0628	<0.01	0.0527	
						20	0.0179	<0.01	0.0273	<0.0552, <0.0934 (<0.0493)
							0.0128	<0.01	0.0206	
Castroville, CA; 2014 (CA65)	10	Artichoke/ Green globe perennial	Flower head	Closer SC	0.272 [306.4]	3	0.293	<0.01	0.0203	<0.323, <0.337 (<0.330)
							0.227	<0.01	0.0997	
L'Acadie, QC; 2014 (QC419)	5B	Artichoke/ Imperial Star	Flower head	Closer SC	0.271 [302.5]	3	0.234	<0.01	0.0824	<0.326, <0.277 (<0.302)
							0.199	<0.01	0.0680	

[1] Expressed as parent equivalents. To express the metabolite residues as parent equivalents, residues of each metabolite were multiplied by the ratio of the molecular weights of sulfoxaflor and the respective metabolite. Therefore residues of X11719474 were multiplied by 277.27/295.29, and residues of X11721061 were multiplied by 277.27/191.15.
[2] Values < LOQ are assumed to be at the LOQ (0.01 ppm).
[3] Total = Sulfoxaflor + X11719474 + X11721061.

Table B.7.6.1.1-7. Summary of Residues from Globe Artichoke Field Trials with Sulfoxaflor.

Crop Matrix	Analyte	Total Application Rate (lbs a.i./A) [g a.i./ha]	PHI (days)	n	Residues[1] (ppm)					
					Max.[2]	LAFT[3]	HAFT[3]	Median[3]	Mean[3]	SD[3]
Globe artichoke flower heads	Sulfoxaflor	0.269-0.272 [301.1-306.4]	3	3	0.293	0.179	0.260	0.217	0.218	0.0408
	X11719474	0.269-0.272 [301.1-306.4]	3	3	<0.01	<0.01	<0.01	<0.01	<0.01	--
	X11721061	0.269-0.272 [301.1-306.4]	3	3	0.0997	0.0595	0.0752	0.0600	0.0649	0.00893
	Total[4]	0.269-0.272 [301.1-306.4]	3	3	<0.337	<0.248	<0.330	<0.302	<0.293	0.0417

n = number of independent field trials, LAFT = lowest average field trial, HAFT = highest average field trial, SD = standard deviation

[1] Expressed as parent equivalents.
[2] Values based on total number of samples.
[3] Values based on per-trial averages.
[4] Total = Sulfoxaflor + X11719474 + X11721061.

Note 1: For computation of the LAFT, HAFT, median, mean, and standard deviation, values < LOQ are assumed to be at the LOQ (0.01 ppm).

III. CONCLUSIONS

The globe artichoke field trials are considered scientifically acceptable. The results of the study showed that following a total application of 0.272 lbs a.i./A (306.4 g a.i./ha) in globe artichoke samples collected at PHIs of 3 days, average combined residues of sulfoxaflor, X11719474, and X11721061 ranged from <0.248 ppm to <0.330 ppm. A decline study indicates that the level of residues in globe artichokes decreases with time. Adequate storage stability data are available to support sample storage durations and conditions.

REFERENCE

PMRA # 1941241. MRID # 47832031. Rodrigues Junior, A. (2010) "Enforcement Method for the Determination of Sulfoxaflor (XDE-208) and its Main Metabolites in Agricultural Commodities using Offline Solid-Phase Extraction and Liquid Chromatography with Tandem Mass Spectrometry Detection." Laboratory Study ID: 091116. Unpublished study prepared by Dow AgroSciences Ind. Ltda, Mogi Mirim, SP. 93 pages.